

1 We claim:

2 1. A composition comprising:

3 an siRNA or shRNA targeted to a target transcript, wherein the target
4 transcript is an agent-specific transcript, which transcript is involved in infection by
5 or replication of an infectious agent.

6 2. The composition of claim 1, wherein:

7 the infectious agent is an agent whose genome comprises multiple
8 independent nucleic acid molecules.

9 3. The composition of claim 2, wherein:

10 the nucleic acid molecules are RNA.

11 4. The composition of claim 2, wherein:

12 the RNA molecules are single-stranded.

13 5. The composition of claim 1, wherein:

14 multiple variants of the infectious agent exist and wherein the agent is
15 capable of undergoing genetic reassortment.

16 6. The composition of claim 1, wherein:

17 multiple variants of the infectious agent exist and wherein the siRNA or
18 shRNA comprises a duplex region whose antisense strand or antisense portion is
19 perfectly complementary to a portion of a target mRNA, which portion is at least 10
20 nucleotides in length and is highly conserved among a plurality of variants.

21 7. The composition of claim 6, wherein:

22 multiple variants of the infectious agent exist and wherein the siRNA or
23 shRNA comprises a duplex region whose antisense strand or antisense portion is
24 perfectly complementary to a portion of a target mRNA, which portion is at least 12
25 nucleotides in length and is highly conserved among a plurality of variants.

26 8. The composition of claim 6, wherein:

1 multiple variants of the infectious agent exist and wherein the siRNA or
2 shRNA comprises a duplex region whose antisense strand or antisense portion is
3 perfectly complementary to a portion of a target mRNA, which portion is at least 15
4 nucleotides in length and is highly conserved among a plurality of variants.

5 9. The composition of claim 6, wherein:

6 multiple variants of the infectious agent exist and wherein the siRNA or
7 shRNA comprises a duplex region whose antisense strand or antisense portion is
8 perfectly complementary to a portion of a target mRNA, which portion is at least 17
9 nucleotides in length and is highly conserved among a plurality of variants.

10 10. The composition of claim 6, wherein:

11 multiple variants of the infectious agent exist and wherein the siRNA or
12 shRNA comprises a duplex region whose antisense strand or antisense portion is
13 perfectly complementary to a portion of a target mRNA, which portion is at least 19
14 nucleotides in length and is highly conserved among a plurality of variants.

15 11. The composition of claim 8, wherein:

16 a portion is highly conserved among variants if it is identical among the
17 different variants.

18 12. The composition of claim 8, wherein

19 a portion is highly conserved among variants if it varies by at most one
20 nucleotide between different variants.

21 13. The composition of claim 8, wherein:

22 a portion is highly conserved among variants if it varies by at most two
23 nucleotides between different variants.

24 14. The composition of claim 8 wherein:

25 the portion is highly conserved among at least 5 variants.

26 15. The composition of claim 8, wherein:

27 the portion is highly conserved among at least 10 variants.

28 16. The composition of claim 8, wherein:

1 the infectious agent infects a host cell and the siRNA or shRNA is present at
2 a level sufficient to inhibit production of the agent by a host cell by at least about 50
3 fold.

4 27. The composition of claim 1, wherein:
5 the infectious agent infects a host cell and the siRNA or shRNA is present at
6 a level sufficient to inhibit production of the agent by a host cell by at least about 100
7 fold.

8 28. The composition of claim 1, wherein:
9 the infectious agent infects a host cell and the siRNA or shRNA is present at
10 a level sufficient to inhibit production of the agent by a host cell by at least about 200
11 fold.

12 29. The composition of claim 1, wherein:
13 the target transcript encodes a subunit of a viral RNA polymerase.

14 30. The composition of claim 1, wherein:
15 the target transcript encodes a hemagglutinin or a neuraminidase.

16 31. The composition of claim 1, wherein:
17 the infectious agent is an influenza virus and the target transcript encodes a
18 protein selected from the group consisting of hemagglutinin, neuraminidase,
19 membrane protein 1, membrane protein 2, nonstructural protein 1, nonstructural
20 protein 2, polymerase protein PB1, polymerase protein PB2, polymerase protein PA,
21 polymerase protein NP.

22 32. The composition of claim 1, wherein:
23 the siRNA or shRNA is present at a level sufficient to inhibit replication of
24 the infectious agent.

25 33. The composition of claim 1, wherein:
26 the siRNA or shRNA comprises a base-paired region at least 15 nucleotides
27 long.

28 34. The composition of claim 1, wherein:

the siRNA or shRNA comprises a base-paired region approximately 19 nucleotides long.

3 35. The composition of claim 1, wherein:
4 the siRNA or shRNA comprises a base-paired region at least 15 nucleotides
5 long and at least one single-stranded 3 prime overhang.

6 36. The composition of claim 1, wherein:
7 the siRNA or shRNA comprises a portion that is perfectly complementary to
8 a region of the target transcript, wherein the portion is at least 15 nucleotides in
9 length.

10 37. The composition of claim 1, wherein:
11 the siRNA or shRNA comprises a portion that is perfectly complementary to
12 a portion of the target transcript, with the exception of at most one nucleotide,
13 wherein the portion is at least 15 nucleotides in length.

14 38. The composition of claim 1, wherein:
15 the siRNA or shRNA comprises a portion that is perfectly complementary
16 with a portion of the target transcript with the exception at most two nucleotides,
17 wherein the portion is at least 15 nucleotides in length.

18 39. The composition of claim 1, wherein:
19 the siRNA or shRNA comprises a core duplex region, wherein the sequence
20 of the sense strand or portion of the core duplex region comprises at least 10
21 consecutive nucleotides as set forth in nucleotides 3 through 21 of the sequence
22 presented in any of SEQ ID NOS: 1 through 68.

23 40. The composition of claim 1, wherein:
24 the siRNA or shRNA comprises a core duplex region, wherein the sequence
25 of the sense strand or portion of the core duplex region comprises at least 12
26 consecutive nucleotides as set forth in nucleotides 3 through 21 of the sequence
27 presented in any of SEQ ID NOS: 1 through 68.

28 41. The composition of claim 1, wherein:

1 the siRNA or shRNA comprises a core duplex region, wherein the sequence
2 of the sense strand or portion of the core duplex region comprises at least 15
3 consecutive nucleotides as set forth in nucleotides 3 through 21 of the sequence
4 presented in any of SEQ ID NOS: 1 through 68.

5 42. The composition of claim 1, wherein:

6 the siRNA or shRNA comprises a core duplex region, wherein the sequence
7 of the sense strand or portion of the core duplex region comprises at least 17
8 consecutive nucleotides as set forth in nucleotides 3 through 21 of the sequence
9 presented in any of SEQ ID NOS: 1 through 68.

10 43. The composition of claim 1, wherein:

11 the siRNA or shRNA comprises a core duplex region, wherein the sequence
12 of the sense strand or portion of the core duplex region comprises at least 19
13 consecutive nucleotides as set forth in nucleotides 3 through 21 of the sequence
14 presented in any of SEQ ID NOS: 1 through 68.

15 44. The composition of claim 1, wherein:

16 the siRNA or shRNA comprises a core duplex region, wherein the sequence
17 of the sense strand or portion of the core duplex region comprises at least 10
18 consecutive nucleotides as set forth in nucleotides 3 through 21 of the sequence
19 presented in any of SEQ ID NOS: 1 through 68, with the proviso that either one or
20 two nucleotides among the 10 consecutive nucleotides may differ from that
21 sequence.

22 45. The composition of claim 1, wherein:

23 the siRNA or shRNA comprises a core duplex region, wherein the sequence
24 of the sense strand or portion of the core duplex region comprises at least 12
25 consecutive nucleotides as set forth in nucleotides 3 through 21 of the sequence
26 presented in any of SEQ ID NOS: 1 through 68, with the proviso that either one or
27 two nucleotides among the 12 consecutive nucleotides may differ from that
28 sequence.

29 46. The composition of claim 1, wherein:

1 the siRNA or shRNA comprises a core duplex region, wherein the sequence
2 of the sense strand or portion of the core duplex region comprises at least 15
3 consecutive nucleotides as set forth in nucleotides 3 through 21 of the sequence
4 presented in any of SEQ ID NOS: 1 through 68, with the proviso that either one or
5 two nucleotides among the 15 consecutive nucleotides may differ from that
6 sequence.

7 47. The composition of claim 1, wherein:

8 the siRNA or shRNA comprises a core duplex region, wherein the sequence
9 of the sense strand or portion of the core duplex region comprises at least 17
10 consecutive nucleotides as set forth in nucleotides 3 through 21 of the sequence
11 presented in any of SEQ ID NOS: 1 through 68, with the proviso that either one or
12 two nucleotides among the 17 consecutive nucleotides may differ from that
13 sequence.

14 48. The composition of claim 1, wherein:

15 the siRNA or shRNA comprises a core duplex region, wherein the sequence
16 of the sense strand or portion of the core duplex region comprises at least 19
17 consecutive nucleotides as set forth in nucleotides 3 through 21 of the sequence
18 presented in any of SEQ ID NOS: 1 through 68, with the proviso that either one or
19 two nucleotides among the 19 consecutive nucleotides may differ from that
20 sequence.

21 49. The composition of claim 1, wherein:

22 the siRNA or shRNA comprises a core duplex region, wherein the sequence
23 of the sense strand or portion of the core duplex region comprises at least 10
24 consecutive nucleotides as set forth in nucleotides 1 through 19 of the sequence
25 presented in any of SEQ ID NOS: 190, 192, 194, 196, 198, 200, 202, 204, 206, 208,
26 210, 212, 214, 216, 218, 220, 222, 224, 226, 228, 230, 232, 234, 236, 238, 240, 242,
27 244, 246, 248, 250, 252, 254, 256, 258, 260, 262, 264, 266, or 268.

28 50. The composition of claim 1, wherein:

29 the siRNA or shRNA comprises a core duplex region, wherein the sequence
30 of the sense strand or portion of the core duplex region comprises at least 12
31 consecutive nucleotides as set forth in nucleotides 1 through 19 of the sequence

1 presented in any of SEQ ID NOS: 190, 192, 194, 196, 198, 200, 202, 204, 206, 208,
2 210, 212, 214, 216, 218, 220, 222, 224, 226, 228, 230, 232, 234, 236, 238, 240, 242,
3 244, 246, 248, 250, 252, 254, 256, 258, 260, 262, 264, 266, or 268.

4 51. The composition of claim 1, wherein:
5 the siRNA or shRNA comprises a core duplex region, wherein the sequence
6 of the sense strand or portion of the core duplex region comprises at least 15
7 consecutive nucleotides as set forth in nucleotides 1 through 19 of the sequence
8 presented in any of SEQ ID NOS: 190, 192, 194, 196, 198, 200, 202, 204, 206, 208,
9 210, 212, 214, 216, 218, 220, 222, 224, 226, 228, 230, 232, 234, 236, 238, 240, 242,
10 244, 246, 248, 250, 252, 254, 256, 258, 260, 262, 264, 266, or 268.

11 52. The composition of claim 1, wherein:
12 the siRNA or shRNA comprises a core duplex region, wherein the sequence
13 of the sense strand or portion of the core duplex region comprises at least 17
14 consecutive nucleotides as set forth in nucleotides 1 through 19 of the sequence
15 presented in any of SEQ ID NOS: 190, 192, 194, 196, 198, 200, 202, 204, 206, 208,
16 210, 212, 214, 216, 218, 220, 222, 224, 226, 228, 230, 232, 234, 236, 238, 240, 242,
17 244, 246, 248, 250, 252, 254, 256, 258, 260, 262, 264, 266, or 268.

18 53. The composition of claim 1, wherein:
19 the siRNA or shRNA comprises a core duplex region, wherein the sequence
20 of the sense strand or portion of the core duplex region comprises at least 19
21 consecutive nucleotides as set forth in nucleotides 1 through 19 of the sequence
22 presented in any of SEQ ID NOS: 190, 192, 194, 196, 198, 200, 202, 204, 206, 208,
23 210, 212, 214, 216, 218, 220, 222, 224, 226, 228, 230, 232, 234, 236, 238, 240, 242,
24 244, 246, 248, 250, 252, 254, 256, 258, 260, 262, 264, 266, or 268.

25 54. The composition of claim 1, wherein:
26 the siRNA or shRNA comprises a core duplex region, wherein the sequence
27 of the sense strand or portion of the core duplex region comprises at least 10
28 consecutive nucleotides as set forth in nucleotides 1 through 19 of the sequence
29 presented in any of SEQ ID NOS: 190, 192, 194, 196, 198, 200, 202, 204, 206, 208,
30 210, 212, 214, 216, 218, 220, 222, 224, 226, 228, 230, 232, 234, 236, 238, 240, 242,
31 244, 246, 248, 250, 252, 254, 256, 258, 260, 262, 264, 266, or 268, with the proviso

1 that either one or two nucleotides among the 10 consecutive nucleotides may differ
2 from that sequence.

3 55. The composition of claim 1, wherein:
4 the siRNA or shRNA comprises a core duplex region, wherein the sequence
5 of the sense strand or portion of the core duplex region comprises at least 12
6 consecutive nucleotides as set forth in nucleotides 1 through 19 of the sequence
7 presented in any of SEQ ID NOS: 190, 192, 194, 196, 198, 200, 202, 204, 206, 208,
8 210, 212, 214, 216, 218, 220, 222, 224, 226, 228, 230, 232, 234, 236, 238, 240, 242,
9 244, 246, 248, 250, 252, 254, 256, 258, 260, 262, 264, 266, or 268, with the proviso
10 that either one or two nucleotides among the 12 consecutive nucleotides may differ
11 from that sequence.

12 56. The composition of claim 1, wherein:
13 the siRNA or shRNA comprises a core duplex region, wherein the sequence
14 of the sense strand or portion of the core duplex region comprises at least 15
15 consecutive nucleotides as set forth in nucleotides 1 through 19 of the sequence
16 presented in any of SEQ ID NOS: 190, 192, 194, 196, 198, 200, 202, 204, 206, 208,
17 210, 212, 214, 216, 218, 220, 222, 224, 226, 228, 230, 232, 234, 236, 238, 240, 242,
18 244, 246, 248, 250, 252, 254, 256, 258, 260, 262, 264, 266, or 268, with the proviso
19 that either one or two nucleotides among the 15 consecutive nucleotides may differ
20 from that sequence.

21 57. The composition of claim 1, wherein:
22 the siRNA or shRNA comprises a core duplex region, wherein the sequence
23 of the sense strand or portion of the core duplex region comprises at least 17
24 consecutive nucleotides as set forth in nucleotides 1 through 19 of the sequence
25 presented in any of SEQ ID NOS: 190, 192, 194, 196, 198, 200, 202, 204, 206, 208,
26 210, 212, 214, 216, 218, 220, 222, 224, 226, 228, 230, 232, 234, 236, 238, 240, 242,
27 244, 246, 248, 250, 252, 254, 256, 258, 260, 262, 264, 266, or 268, with the proviso
28 that either one or two nucleotides among the 17 consecutive nucleotides may differ
29 from that sequence.

30 58. The composition of claim 1, wherein:

1 the siRNA or shRNA comprises a core duplex region, wherein the sequence
2 of the sense strand or portion of the core duplex region comprises at least 19
3 consecutive nucleotides as set forth in nucleotides 1 through 19 of the sequence
4 presented in any of SEQ ID NOS: 190, 192, 194, 196, 198, 200, 202, 204, 206, 208,
5 210, 212, 214, 216, 218, 220, 222, 224, 226, 228, 230, 232, 234, 236, 238, 240, 242,
6 244, 246, 248, 250, 252, 254, 256, 258, 260, 262, 264, 266, or 268, with the proviso
7 that either one or two nucleotides among the 19 consecutive nucleotides may differ
8 from that sequence.

9 59. The composition of claim 1, wherein the siRNA or shRNA comprises sense and
10 antisense strands or portions whose sequences comprise sequences given by
11 nucleotides 1 – 19 of SEQ ID NOS: 77 and 78 respectively, with, optionally, a 3'
12 overhang on one or both sequences.

13 60. The composition of claim 1, wherein the siRNA or shRNA comprises sense and
14 antisense portions whose sequences comprise sequences given by nucleotides 1 – 19
15 of SEQ ID NOS: 71 and 72 respectively, with, optionally, a 3' overhang on one or
16 both sequences.

17 61. The composition of claim 1, wherein the siRNA or shRNA comprises sense and
18 antisense portions whose sequences comprise sequences given by nucleotides 1 – 19
19 of SEQ ID NOS: 83 and 84 respectively, with, optionally, a 3' overhang on one or
20 both sequences.

21 62. The composition of claim 1, wherein the siRNA or shRNA comprises sense and
22 antisense portions whose sequences comprise sequences given by nucleotides 1 – 19
23 of SEQ ID NOS: 89 and 90 respectively, with, optionally, a 3' overhang on one or
24 both sequences.

25 63. The composition of claim 1, wherein the siRNA or shRNA comprises sense and
26 antisense portions whose sequences comprise sequences given by nucleotides 1 – 19
27 of SEQ ID NOS: 91 and 92 respectively, with, optionally, a 3' overhang on one or
28 both sequences.

- 1 64. The composition of claim 1, wherein the siRNA or shRNA comprises sense and
2 antisense portions whose sequences comprise sequences given by nucleotides 1 – 19
3 of SEQ ID NOS: 93 and 94 respectively, with, optionally, a 3' overhang on one or
4 both sequences.
- 5 65. The composition of claim 1, wherein the siRNA or shRNA comprises sense and
6 antisense portions whose sequences comprise sequences given by nucleotides 1 – 20
7 of SEQ ID NOS: 188 and 189 respectively, with, optionally, a 3' overhang on one or
8 both sequences.
- 9 66. The composition of claim 1, wherein the siRNA or shRNA comprises a duplex
10 portion selected from the group consisting of duplex portions of: NP-1496, NP-
11 1496a, PA-2087, PB1-2257, PB1-129, PB2-2240, M-37, or M-598 or a variant of
12 any of the foregoing, which variant differs by at most one nucleotide from the
13 corresponding siRNA.
- 14 67. The composition of claim 66, wherein the siRNA or shRNA duplex portion is
15 identical to the duplex portion of NP-1496.
- 16 68. The composition of claim 66, wherein the siRNA or shRNA duplex portion is
17 identical to the duplex portion of NP-1496a.
- 18 69. The composition of claim 1, wherein the sense strand or portion of the siRNA or
19 shRNA has a sequence selected from the group consisting of: the first 19 nucleotides
20 of SEQ ID NO: 71, SEQ ID NO: 75, SEQ ID NO: 77, SEQ ID NO: 83, SEQ ID NO:
21 93; SEQ ID NO: 95; SEQ ID NO: 99, and SEQ ID NO: 188, reading in a 5' to 3'
22 direction.
- 23 70. An analog of the siRNA or shRNA of claim 1, wherein the analog differs from the
24 siRNA or shRNA in that it contains at least one modification.
- 25 71. The analog of claim 70, wherein:
26 the modification results in increased stability of the siRNA, enhances
27 absorption of the siRNA, enhances cellular entry of the siRNA, or any combination
28 of the foregoing.

- 1 72. The analog of claim 70, wherein:
 - 2 the modification modifies a base, a sugar, or an internucleoside linkage.
- 3 73. The analog of claim 70, wherein:
 - 4 the modification is not a nucleotide 2' modification.
- 5 74. The analog of claim 70, wherein:
 - 6 the modification is a nucleotide 2' modification.
- 7 75. An analog of the siRNA or shRNA of claim 1, wherein:
 - 8 the analog differs from the siRNA in that at least one ribonucleotide is
 - 9 replaced by a deoxyribonucleotide.
- 10 76. A composition comprising a plurality of single-stranded RNAs which, when
11 hybridized to each other, form the composition of claim 1.
- 12 77. The composition of claim 76, wherein:
 - 13 the single-stranded RNAs range in length between approximately 21 and 23
 - 14 nucleotides, inclusive.
- 15 78. A composition comprising a plurality of the siRNAs or shRNAs of claim 1.
- 16 79. The composition of claim 78, wherein at least some of the siRNAs or shRNAs are
17 targeted to different influenza virus transcripts.
- 18 80. The composition of claim 78, wherein at least some of the siRNAs or shRNAs are
19 targeted to different regions of the same influenza virus transcript.
- 20 81. The siRNA or shRNA of claim 1, wherein:
 - 21 presence of the siRNA or shRNA within a cell susceptible to influenza virus
 - 22 infection reduces the susceptibility of the cell to infection by at least two influenza
 - 23 strains.
- 24 82. The siRNA or shRNA of claim 1, wherein presence of the siRNA or shRNA within a
25 subject susceptible to infection with influenza virus reduces the susceptibility of the
- 26 subject to infection by at least two influenza strains.
- 27 83. A cell comprising the siRNA or shRNA of claim 1.

- 1 84. A vector that provides a template for synthesis of the siRNA or shRNA of claim 1.
- 2 85. The vector of claim 84, wherein the vector comprises a nucleic acid operably linked
3 to expression signals active in a host cell so that, when the construct is introduced
4 into the host cell, the siRNA or shRNA of claim 1 is produced inside the host cell
- 5 86. A vector comprising a nucleic acid operably linked to expression signals active in a
6 host cell so that, when the construct is introduced into the host cell, an siRNA or
7 shRNA is produced inside the host cell that is targeted to an transcript specific to an
8 infectious agent, which transcript is involved in infection by or replication of the
9 agent.
- 10 87. The vector of claim 86, wherein the infectious agent is a virus and wherein multiple
11 variants of the virus exist and wherein the virus is capable of undergoing genetic
12 reassortment or mixing.
- 13 88. A cell comprising the vector of claim 87.
- 14 89. A transgenic animal comprising the vector of claim 87.
- 15 90. The vector of claim 87, wherein the virus is one whose genome comprises multiple
16 independent nucleic acid molecules.
- 17 91. The vector of claim 87, wherein the infectious agent is an influenza virus.
- 18 92. The vector of claim 91, wherein the vector provides a template for transcription of
19 one or more strands of an siRNA or an shRNA that reduces susceptibility of the cell
20 to infection by influenza virus or inhibits influenza virus production.
- 21 93. The vector of claim 91, wherein degradation of the target transcript delays, prevents,
22 or inhibits one or more aspects of influenza virus infection or replication.
- 23 94. The vector of claim 92, wherein the siRNA or shRNA duplex portion is selected
24 from the group consisting of duplex portions of: NP-1496, NP-1496a, PA-2087,
25 PB1-2257, PB1-129, PB2-2240, M-37, and M-598, or a variant of any of the
26 foregoing, wherein the variant differs by at most one nucleotide from the
27 corresponding siRNA in either its sense portion, antisense portion, or both.

- 1 95. The vector of claim 94, wherein the siRNA or shRNA duplex portion is identical to
- 2 the duplex portion of NP-1496.
- 3 96. The vector of claim 94, wherein the siRNA duplex portion is identical to the duplex
- 4 portion of NP-1496a.
- 5 97. The vector of claim 94, wherein the sense strand or portion of the siRNA or shRNA
- 6 has a sequence selected from the group consisting of: the first 19 nucleotides of any
- 7 of SEQ ID NOS: 71, 75, 77, 83, 93, 95, 99, and 188, reading in a 5' to 3' direction.
- 8 98. The vector of claim 86, wherein:
9 the nucleic acid is operably linked to a promoter for RNA polymerase III.
- 10 99. The vector of claim 98, wherein:
11 the promoter is a U6 or H1 promoter.
- 12 100. The vector of claim 86, wherein:
13 the vector is selected from the group consisting of retroviral vectors,
14 lentiviral vectors, adenovirus vectors, and adeno-associated virus vectors.
- 15 101. The vector of claim 86, wherein the vector is a lentiviral vector.
- 16 102. The vector of claim 86, wherein the vector is a DNA vector.
- 17 103. The vector of claim 86, wherein the vector is a virus.
- 18 104. The vector of claim 86, wherein the vector is a lentivirus.
- 19 105. A method of treating or preventing infection by an infectious agent, the method
20 comprising steps of: administering to a subject prior to, simultaneously with, or after
21 exposure of the subject to the infectious agent, a composition comprising the vector
22 of claim 86 or the cell of claim 88.
- 23 106. The method of claim 105, wherein the infectious agent is a virus.
- 24 107. The method of claim 105, wherein the infectious agent infects respiratory epithelial
25 cells.

- 1 108. The method of claim 105, wherein the infectious agent is an influenza virus.
- 2 109. The method of claim 105, wherein the composition is administered intravenously.
- 3 110. The method of claim 105, wherein the composition is administered intranasally.
- 4 111. The method of claim 105, wherein the composition is administered by inhalation.
- 5 112. A pharmaceutical composition comprising:
 - 6 the composition of claim 1; and
 - 7 a pharmaceutically acceptable carrier.
- 8 113. The pharmaceutical composition of claim 112, wherein:
 - 9 the composition is formulated as an aerosol.
- 10 114. The pharmaceutical composition of claim 112, wherein:
 - 11 the composition is formulated as a nasal spray.
- 12 115. The pharmaceutical composition of claim 112, wherein:
 - 13 the composition is formulated for intravenous administration.
- 14 116. The pharmaceutical composition of claim 112, wherein:
 - 15 the infectious agent is an influenza virus and wherein the composition further
 - 16 comprises a second anti-influenza agent.
- 17 117. The pharmaceutical composition of claim 116, wherein the second anti-influenza
- 18 agent is approved by the United States Food and Drug Administration.
- 19 118. A method for identifying viral inhibitors, the method comprising steps of:
 - 20 providing a cell including a candidate siRNA or shRNA whose sequence
 - 21 includes a region of complementarity with at least one transcript produced during
 - 22 infection with a virus, which transcript is characterized in that its degradation delays,
 - 23 prevents, or inhibits one or more aspects of viral infection or replication;
 - 24 detecting infection by or replication of the virus in the cell; and
 - 25 identifying an siRNA or shRNA that inhibits viral infectivity or replication,
 - 26 which siRNA or shRNA is a viral inhibitor.
- 27 119. The method of claim 118, wherein:

1 the virus is an influenza virus.

2 120. The method of claim 118, wherein:

3 the cell is characterized in that in the absence of the siRNA or shRNA the cell
4 produces at least one viral transcript.

5 121. The method of claim 118, further comprising the step of:

6 transfecting the cell with a viral genome or infecting the cell with the virus.

7 122. A method of treating or preventing infection by a virus, the method comprising steps
8 of:

9 administering to a subject prior to, simultaneously with, or after exposure of
10 the subject to the virus, a composition comprising an effective amount of an RNAi-
11 inducing entity, wherein the RNAi-inducing entity is targeted to a transcript
12 produced during infection by the virus, which transcript is characterized in that
13 reduction in levels of the transcript delays, prevents, or inhibits one or more aspects
14 of infection by or replication of the virus.

15 123. The method of claim 122, wherein:

16 the virus infects respiratory epithelial cells.

17 124. The method of claim 122, wherein:

18 the virus is an influenza virus.

19 125. The method of claim 122, wherein the composition is administered into the
20 respiratory tract.

21 126. The method of claim 122, wherein the composition is administered by a conventional
22 intravenous delivery method.

23 127. The method of claim 122, wherein in the absence of the RNAi-inducing entity the
24 virus is able to undergo a complete life cycle leading to production of infectious
25 virus, and wherein the presence of the siRNA or shRNA inhibits production of the
26 virus.

27 128. The method of claim 122, wherein the RNAi-inducing entity comprises a duplex
28 portion selected from the group consisting of: duplex portions of: NP-1496, NP-

1 1496a, PA-2087, PB1-2257, PB1-129, PB2-2240, M-37, and M-598, or a variant of
2 any of the foregoing, wherein the variant differs by at most one nucleotide from the
3 corresponding siRNA in either its sense portion, antisense portion, or both.

4 129. The method of claim 128, wherein the duplex portion is identical to the duplex
5 portion of NP-1496.

6 130. The vector of claim 128, wherein the duplex portion is identical to the duplex portion
7 of NP-1496a.

8 131. A method for designing an siRNA or shRNA having a duplex portion, the method
9 comprising steps of:
10 identifying a portion of a target transcript, which portion is highly conserved
11 among a plurality of variants of an infectious agent and comprises at least 15
12 consecutive nucleotides; and
13 selecting the sequence of the portion as the sequence for the duplex portion of
14 the siRNA or shRNA sense strand or portion.

15 132. The method of claim 131, further comprising:
16 selecting a sequence complementary to the portion as the sequence for the
17 duplex portion of the siRNA or shRNA antisense strand or portion.

18 133. The method of claim 132, further comprising:
19 adding a 3' overhang to either or both of the sense and antisense strands of
20 the siRNA duplex.

21 134. The method of claim 131, wherein:
22 the plurality of variants comprises at least 10 variants.

23 135. The method of claim 131, wherein:
24 the plurality of variants comprises at least 15 variants.

25 136. The method of claim 131, wherein:
26 the plurality of variants comprises at least 20 variants.

27 137. The method of claim 131, wherein:
28 the portion comprises approximately 19 nucleotides.

- 1 138. The method of claim 131, wherein:
 - 2 a portion is considered highly conserved among a plurality of variants if it
 - 3 differs by at most one nucleotide between the variants.
- 4 139. The method of claim 131, wherein:
 - 5 the infectious agent is an influenza virus.
- 6 140. The method of claim 131, wherein:
 - 7 the infectious agent is capable of undergoing reassortment.
- 8 141. The method of claim 131, wherein:
 - 9 the variants include at least two variants, each of which naturally infects a
 - 10 host of a different species.
- 11 142. The method of claim 141, wherein:
 - 12 the species include at least two species selected from the group consisting of
 - 13 humans, swine, horse, and bird species.
- 14 143. The method of claim 131, wherein:
 - 15 the variants include at least two variants, each of which arose in a host of a
 - 16 different species.
- 17 144. The method of claim 143, wherein:
 - 18 the species include at least two species selected from the group consisting of
 - 19 humans, swine, horse, and bird species.
- 20 145. A composition comprising an siRNA or shRNA designed in accordance with the
- 21 method of claim 131.
- 22 146. A method of reducing or lowering levels of a transcript, which transcript is a vRNA
- 23 or cRNA, comprising administering an RNAi-inducing entity targeted to an mRNA
- 24 transcript having a sequence at least a portion of which is complementary to or
- 25 identical to the vRNA or cRNA transcript.
- 26 147. A method of inhibiting a first transcript comprising administering an RNAi-inducing
- 27 entity targeted to a second transcript, wherein inhibition of the second transcript
- 28 results in inhibition of the first transcript.

- 1 148. The method of claim 147, wherein the level of the first transcript is reduced relative
- 2 to its level in the absence of the RNAi-inducing entity.
- 3 149. The method of claim 147, wherein the level of the second transcript is reduced
- 4 relative to its level in the absence of the RNAi-inducing entity.
- 5 150. The method of claim 147, wherein the levels of the first and second transcript are
- 6 reduced relative to their levels in the absence of the RNAi-inducing entity.
- 7 151. The method of claim 147, wherein the RNAi-inducing entity is not specifically
- 8 targeted to the first transcript.
- 9 152. The method of claim 147, wherein the second transcript encodes a protein that
- 10 functions in maintaining RNA stability.
- 11 153. The method of claim 147, wherein the protein is a nucleic acid binding protein.
- 12 154. The method of claim 153, wherein the nucleic acid binding protein is an RNA
- 13 binding protein.
- 14 155. The method of claim 147, wherein the second transcript encodes a polymerase.
- 15 156. The method of claim 155, wherein the polymerase is an RNA polymerase.
- 16 157. The method of claim 155, wherein the polymerase is a DNA polymerase.
- 17 158. The method of claim 155, wherein the polymerase is a reverse transcriptase.
- 18 159. The method of claim 147, wherein either of both of the first and second transcripts
- 19 are agent-specific transcripts, wherein the agent is an infectious agent.
- 20 160. The method of claim 147, wherein the first and second transcripts are agent-specific
- 21 transcripts, wherein the agent is an infectious agent.
- 22 161. The method of claim 160, wherein the infectious agent is a virus.
- 23 162. The method of claim 161, wherein the virus is an influenza virus.

1 163. The method of claim 162, wherein the second transcript encodes either viral NP
2 protein or viral PA protein.

3 164. The method of claim 163, wherein the first transcript encodes a protein selected from
4 the group consisting of: M protein, HA protein, PB1 protein, PB2 protein, or NS
5 protein.

6 165. A composition comprising:
7 an RNAi-inducing entity, wherein the RNAi-inducing entity is targeted to an
8 influenza virus transcript; and
9 a delivery agent selected from the group consisting of: cationic polymers,
10 modified cationic polymers, peptide molecular transporters, surfactants suitable for
11 introduction into the lung, neutral or cationic lipids, liposomes, non-cationic
12 polymers, modified non-cationic polymers, bupivacaine, and chloroquine.

13 166. The composition of claim 165, wherein the delivery agent comprises a delivery-
14 enhancing moiety to enhance delivery to a cell of interest.

15 167. The composition of claim 165, wherein the delivery-enhancing moiety comprises an
16 antibody, antibody fragment, or ligand that specifically binds to a molecule
17 expressed by the cell of interest.

18 168. The composition of claim 167, wherein the cell of interest is a respiratory epithelial
19 cell.

20 169. The composition of claim 165, wherein the delivery-enhancing moiety comprises a
21 moiety selected to reduce degradation, clearance, or nonspecific binding of the
22 delivery agent.

23 170. The composition of claim 165, wherein the RNAi-inducing entity comprises a viral
24 vector.

25 171. The composition of claim 170, wherein the viral vector comprises a lentiviral vector.

26 172. The composition of claim 165, wherein the RNAi-inducing entity comprises a DNA
27 vector.

- 1 173. The composition of claim 165, wherein the RNAi-inducing entity comprises a virus.
- 2 174. The composition of claim 173, wherein the RNAi-inducing entity comprises a
3 lentivirus.
- 4 175. The composition of claim 165, wherein the RNAi-inducing entity comprises an
5 siRNA.
- 6 176. The composition of claim 165, wherein the RNAi-inducing entity comprises an
7 shRNA.
- 8 177. The composition of claim 165, wherein the RNAi-inducing entity comprises an
9 RNAi-inducing vector whose presence within a cell results in production of an
10 siRNA or shRNA targeted to an influenza virus transcript.
- 11 178. The composition of claim 165, wherein:
12 the RNAi-inducing entity comprises an siRNA or shRNA or an RNAi-
13 inducing vector whose presence within a cell results in production of an siRNA or
14 shRNA, wherein the siRNA or shRNA comprises a portion that is perfectly
15 complementary to a region of the target transcript, wherein the portion is at least 15
16 nucleotides in length.
- 17 179. The composition of claim 165, wherein:
18 the RNAi-inducing entity comprises an siRNA or shRNA or an RNAi-
19 inducing vector whose presence within a cell results in production of an siRNA or
20 shRNA, wherein the siRNA or shRNA comprises a duplex portion selected from the
21 group consisting of duplex portions of: NP-1496, NP-1496a, PA-2087, PB1-2257,
22 PB1-129, PB2-2240, M-37, and M-598, or a variant of any of the foregoing, wherein
23 the variant differs by at most one nucleotide from the corresponding siRNA or
24 shRNA in either its sense portion, antisense portion, or both.
- 25 180. The composition of claim 179, wherein the siRNA or shRNA duplex portion
26 comprises the duplex portion of NP-1496.
- 27 181. The composition of claim 179, wherein the siRNA or shRNA duplex portion
28 comprises the duplex portion of NP-1496a.

- 1 182. The composition of claim 165, wherein:
 - 2 the RNAi-inducing entity comprises an siRNA or shRNA or an RNAi-inducing vector whose presence within a cell results in production of an siRNA or shRNA, wherein the siRNA or shRNA, wherein the sequence of the sense strand or portion of the siRNA or shRNA comprises a sequence selected from the group consisting of: the first 19 nucleotides of, SEQ ID NO: 71, SEQ ID NO: 75, SEQ ID NO: 77, SEQ ID NO: 83, SEQ ID NO: 93; SEQ ID NO: 95; SEQ ID NO: 99, and SEQ ID NO: 188 reading in a 5' to 3' direction.
- 9 183. The composition of claim 182, wherein the sequence of the sense strand or portion of the siRNA or shRNA comprises the sequence of SEQ ID NO: 93.
- 11 184. The composition of claim 182, wherein the sequence of the sense strand or portion of the siRNA or shRNA comprises the sequence of SEQ ID NO: 188.
- 13 185. The composition of claim 165, wherein the delivery agent is selected from the group consisting of cationic polymers, modified cationic polymers, and surfactants suitable for introduction into the lung.
- 16 186. The composition of claim 185, wherein the cationic polymer is selected from the group consisting of polylysine, polyarginine, polyethyleneimine, polyvinylpyrrolidone, chitosan, and poly(β -amino ester) polymers.
- 19 187. The composition of claim 186, wherein the cationic polymer is polyethyleneimine.
- 20 188. The composition of claim 185, wherein the modified cationic polymer incorporates a modification selected to reduce the cationic nature of the polymer.
- 22 189. The composition of claim 188, wherein the modification comprises substitution with a group selected from the list consisting of: acetyl, imidazole, succinyl, and acyl.
- 24 190. The composition of claim 185, wherein between 25% and 75% of the residues of the modified cationic polymer are modified.
- 26 191. The composition of claim 190, wherein approximately 50% of the residues of the modified cationic polymer are modified.

- 1 192. The composition of claim 185, wherein the delivery agent comprises a surfactant
2 suitable for introduction into the lung.
- 3 193. The composition of claim 192, wherein the surfactant is Infasurf®, Survanta®, or
4 Exosurf®.
- 5 194. A method of treating or preventing influenza virus replication, pathogenicity, or
6 infectivity comprising administering the composition of claim 165 to a subject at risk
7 of or suffering from influenza virus infection.
- 8 195. The method of claim 194, wherein the composition is administered by a route
9 selected from the group consisting of: intravenous injection, inhalation, intranasally,
10 and as an aerosol.
- 11 196. The method of claim 194, wherein the composition is administered intravenously.
- 12 197. The method of claim 196, wherein the composition is administered using a
13 conventional intravenous administration technique.
- 14 198. The method of claim 194, wherein the composition is administered by inhalation.
- 15 199. The method of claim 194, wherein the composition is administered intranasally.
- 16 200. The method of claim 194, wherein the composition is administered as an aerosol.